

CUTANEOUS TOXICITY EVALUATION OF AIR FORCE DEVELOPMENT MATERIALS - VIII

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INDUSTRIAL BIOLOGY RESEARCH AND TESTING
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FOREWORD

This report was initiated by the Toxic Hazards Branch, Physiology Division, Biomedical Laboratory of the Aerospace Medical Research Laboratories. The contract monitor was Dr. Kenneth C. Back. The original research and development work upon which the report is based was accomplished by Industrial Biology Research and Testing Laboratories, Inc., 22 N. 36th Street, Philadelphia, Pennsylvania under Air Force Contract No. AF 33 (615)-1571, in support of Project No. 6302, "Toxic Hazards of Propellants and Materials," Task No. 630201, "Toxicology." The author, Dr. Morris V. Shelanski, was project director in charge of the basic research and development work. Research was begun in June 1965 and completed in October 1965. Mr. Hyman R. Gittes, Toxicologist, and Dr. Theodore Levenson, Chemist, cooperated in the research.

This is the eighth in a series of reports, entitled "Cutaneous Toxicity Evaluation of Air Force Development Materials," by the Industrial Biology Research and Testing Laboratories, Inc. The previous reports are:

- I. WADC-TR 56-626, December 1956, by
M. V. Shelanski and C. Josephs
- II. WADC TR 57-742, November 1957, by
M. V. Shelanski and K. L. Gabriel
- III. WADC TR 59-124, June 1959, by
M. V. Shelanski and K. L. Gabriel
- IV. ASD TR 61-77, April 1961, by
M. V. Shelanski and K. L. Gabriel
- V. MLR-TDR-62-26, April 1962, by
M. V. Shelanski and K. L. Gabriel
- VI. AMRL-TDR-64-13, February 1964, by
M. V. Shelanski
- VII. AMRL-TR-64-120, December 1964, by
M. V. Shelanski

This technical report has been reviewed and is approved.

WAYNE H. McCANDLESS
Technical Director, Physical Sciences
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ABSTRACT

Four Air Force development materials were studied via the prophetic patch test method on laboratory animals to determine the primary irritant effect, gross sensitization index, and gross percutaneous toxicity of these materials. The patch test studies with rabbits indicated that one of the materials produced severe primary irritant action. Testing on human volunteers was therefore carried out with three of the materials. Results indicated that these materials were safe to use in contact with human skin.

INTRODUCTION

Industrial Biology Research and Testing Laboratories, Inc. was engaged by the United States Air Force to perform dermatological studies and provide cutaneous toxicity data on certain Air Force development materials. These data would serve the Air Force as criteria for establishing safe handling procedures and limits of application of these materials when utilized by Air Force personnel.

There are various methods used for the determination of cutaneous toxicity of a chemical compound or substance. Laboratory animals, such as rabbits or guinea pigs, have been used by many investigators (ref. 1). The true index of cutaneous reaction can, however, only be determined by using human subjects. Prophetic patch tests are one of the methods used for this purpose (refs. 2 & 3). This test method helps to establish both the primary irritation and sensitization characteristics of a compound brought into contact with the human skin. Prophetic patch test studies were performed on laboratory animals to screen the primary irritant and sensitization characteristics of certain Air Force development materials. The Shelanski repeated insult patch test (ref. 3), in addition to giving information about primary irritation and sensitization characteristics of the compound, will also bring out any fatiguing reactions which may occur on continuous contact of the material with the human skin. This technique was performed on volunteer human subjects to define the characteristics of these compounds on the skin of humans.

MATERIALS

The following materials were procured upon instructions from the Aerospace Medical Research Laboratories:

1. Spray disinfectant (Lysol - Lehan & Fink, Bloomfield, N. J.)
2. Para nitrophenol, reagent grade
3. Para nitrophenol, technical grade
4. Tributyl tin-n-octyl succinate as contained in Code No. EC-2241 manufactured by The 3 M Company. Concentration of the organo-tin is 1.9% by weight. The balance consists of volatile components (toluene 25%, xylene 35%, butyl alcohol about 10%) and non-volatile components (pigments, elastomer, titanium dioxide and a vulcanizing agent).

CRITERIA
FOR GRADING PATCH TEST REACTIONS

The investigators have discussed the criteria for grading patch test reactions used by various authors in a previous report, March 1955 (ref. 4). In this study, as in the previous, the following criteria were used by the Industrial Biology Research and Testing Laboratories, Inc.:

- 0 - no reaction, or questionable reaction
- 1+ - definite or clear-cut erythema
- 2+ - marked erythema, greater than present in 1+ reaction
- 3+ - marked erythema, edema, with or without a few vesicles
- 4+ - marked erythema, edema, with vesicles and oozing

RABBIT SCREENING STUDIES

PROCEDURE

Five groups of five albino rabbits each were used in this study. The animals selected weighed approximately two kilograms each. Prior to use, the animals were placed on colony diet and observed for a period of two weeks. Animals not showing normal weight gain were replaced.

Prior to patching, the fur on the back of each rabbit was closely clipped to expose an area of skin equal to at least 10% of the total body area. This area was then shaved to denude the skin completely. The patch site area was marked with permanent ink to identify the site for later reference.

The test materials were used as follows for each application:

The Lysol spray disinfectant was expressed into a glass container and allowed to stand loosely stoppered overnight. Approximately 4 ml of the liquid was spread over the exposed skin, covered with glassine paper and held in place by a muslin binder.

The para nitrophenols, both reagent grade and technical grade, were each made up as a 5% w/v solution in a solvent consisting by volume of 95% butanol and 5% isopropanol. These were painted on sheets of cork approximately 2 mm thick and allowed to dry. Approximately 4 cm² of the cork was placed on each rabbit's shaved skin with painted side in contact with the skin. The cork was held in place by means of a muslin binder and adhesive tape.

The EC-2241 was painted on sheets of cork approximately 2 mm in thickness and allowed to dry. Approximately 4 cm² of the cork was placed on each rabbit's shaved skin with the painted side in contact with the skin. The cork was held in place by means of a muslin binder and adhesive tape.

Five rabbits per material were used. The first or primary application remained in contact with the denuded skin for forty-eight hours. Upon removal, reactions were graded and recorded. Twenty-four hours after removal of the patches, the sites were examined for delayed reactions.

Following the primary application, the animals were rested for fourteen days. The patch material was then reapplied on the same site as a challenge or sensitization application. Again, after forty-eight hours contact, the patches were removed and reactions graded and recorded. Twenty-four hours later, the sites were examined for delayed reactions. In the case of the EC-2241, applications were made to a fresh site. This was necessary by reason of the fact that lesions resulting from the primary applications had not healed sufficiently to permit reapplication to the original site.

RESULTS

Material No. 1 - Spray disinfectant (Lysol - Lehan & Fink, Bloomfield, N. J.) - produced no reactions in any of the five rabbits to either the initial or challenge applications.

Material No. 2 - Para nitrophenol, reagent grade - produced no reactions in any of the five rabbits to either the initial or challenge applications.

Material No. 3 - Para nitrophenol, technical grade - produced no reactions in any of the five rabbits to either the initial or challenge applications.

Material No. 4 - Tributyl tin-n-octyl succinate (The 3 M Company Code No. EC-2241) - produced marked erythema and edema. This was apparent upon removal of the patches. No intensification of these reactions was noted during subsequent observations. At fourteen days following the primary applications the lesions present precluded further use of the site. A similar response pattern to the challenge application was noted:

Rabbit Number	<u>Primary Application</u>				<u>Challenge Application</u>			
	Day				Day			
	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>
1	3+	3+	3+	3+	3+	3+	3+	3+
2	3+	3+	3+	3+	3+	3+	3+	3+
3	3+	3+	3+	3+	3+	3+	3+	3+
4	3+	3+	3+	3+	3+	3+	3+	3+
5	3+	3+	3+	3+	3+	3+	3+	3+

CONCLUSIONS

Effects produced by the EC-2241 were considered of sufficient severity to preclude testing in humans.

HUMAN PATCH TESTS

SHELANSKI REPEATED INSULT PATCH TEST

PROCEDURE

Each material was tested on three hundred human volunteer subjects. For this testing the Lysol spray disinfectant was expressed into a glass container and allowed to stand loosely stoppered overnight; the para nitrophenols were dissolved and painted on cork sheets as described for the rabbit test phase. The materials were applied, using the conventional patch technique, to the skin of the subjects for twenty-four hours and then removed. Skin reactions were graded and recorded. The skin was allowed to recuperate for twenty-four hours. This cycle of contact and recuperation was repeated fifteen times for a total of thirty days, the reaction being graded after each application. Following the removal and the grading of the fifteenth application the skin was allowed to recuperate for two weeks. The material was then re-applied on the same subjects for twenty-four hours. Patches were then removed and the reactions were graded and recorded. The first application gave an index of primary irritation. The final application gave information on sensitization. The repeated applications, from the second through the fifteenth, determined the extent of fatiguing and served to accelerate skin reactions which facilitated forecasting of probability of cutaneous irritation due to long-term exposures.

RESULTS

Material No. 1 - Spray disinfectant (Lysol - Lehan & Fink, Bloomfield, N. J.). This material was not a primary irritant or a fatiguing agent to the 300 human volunteer subjects. This material did not sensitize any of the subjects.

Material No. 2 - Para nitrophenol, reagent grade. This material was not a primary irritant or a fatiguing agent to the 300 human volunteer subjects. This material did not sensitize any of the subjects.

Material No. 3 - Para nitrophenol, technical grade. This material was not a primary irritant or a fatiguing agent to the 300 human volunteer subjects. This material did not sensitize any of the subjects.

CONCLUSIONS

In this study of four materials, three materials, namely, Spray disinfectant (Lysol - Lehan & Fink, Bloomfield, N. J.), Para nitrophenol, reagent grade and Para nitrophenol, technical grade, produced no significant reactions by either the Schwartz prophetic patch test on rabbits (ref. 2) or the Shelanski repeated insult patch test on three hundred human volunteers (ref. 3). These materials may be considered innocuous and may be permitted to contact human skin for prolonged periods. This conclusion is based upon a generally accepted testing procedure. However, it must be pointed out that the test method is not infallible or above criticism. Further, the patch test situation does not duplicate the range of temperature, humidity, air flow, perspiration, and friction, among other factors, which will be met in actual usage of the material. Because the prophetic patch test was devised to provide screening information with respect to cutaneous irritation and sensitivity from certain materials, it must be emphasized that the test should be used only for that purpose. The recommended procedure following the test is to employ the material within the limits recommended for direct skin contact on a usage basis. This should be done on 5,000 to 10,000 subjects, preferably under variable climatic conditions prior to the release of the material for general use.

With respect to the remaining material tested it is concluded that:

Tributyl tin-n-octyl succinate as contained in Code No. EC-2241 manufactured by The 3 M Company, due to its severe effect upon rabbit skin, is not safe to use in contact with the human skin.

REFERENCES

1. Draize, John H.; Woodard, Geoffrey; and Calvery, Herbert O.: "Methods for the Study of Irritation and Toxicity of Substances Applied Topically to the Skin and Mucous Membranes." The Journal of Pharmacology and Experimental Therapeutics, 82: No. 4, 377-390, December 1944.
2. Schwartz, L., and Peck, S. M.: "The Patch Test in Contact Dermatitis." Public Health Reports, 59: 546-557, April 1944. Reprint No. 2552.
3. Shelanski, H. A. and Shelanski, M. V.: "A New Technique of Human Patch Tests." Proceedings of the Scientific Section of the Toilet Goods Association, 19: 46-49, May 1953.
4. Shelanski, Morris V. and Josephs, Charles: Cutaneous Toxicity Evaluation of Fabrics Impregnated with Anti-Mildew Agents, WADC Technical Report 55-198, Wright-Patterson AFB, Ohio, March 1955.

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